**Synopsis**
**of the thesis entitled**

# EFFECT OF EFFICIENT MICROBES ON PLANT GROWTH PROMOTION AND ANTIBACTERIAL ACTIVITY OF *Sphaeranthes amaranthoides* (L.) Burm.

to be submitted to
**Bharathidasan University, Tiruchirappalli**
for the award of the degree of
**DOCTOR OF PHILOSOPHY**
**IN**
**MICROBIOLOGY**

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**SEPTEMBER 2009**

## (A) Title of the Thesis :

**EFFECT OF EFFICIENT MICROBES ON PLANT GROWTH PROMOTION AND ANTIBACTERIAL ACTIVITY OF *Sphaeranthes amaranthoides* (L.) Burm.**

## (B) Brief description on the state of the art of the Research topic:

Health is the real wealth of a nation. Nature has provided all necessary things for our survival. Medicinal plants are nature's best gift to cure a number of diseases of men and animals. From 1980 onwards there was an alarming renewed interest around the world in plant-based drugs. There are about 121 clinically useful prescription drugs that are worldwide derived from higher plants.

Plants have formed the basis for traditional medicine systems that have been used for thousands of years in countries such as China (Chang and But, 1986) and India (Kapoor, 1990). The use of plants in the traditional medicinal systems of many other cultures has been extensively documented. These plant based systems continue to play an essential role in healthcare and it has been estimated by the World Health Organization (WHO), that approximately 80% of the world inhabitants relay mainly on traditional medicines for their primary healthcare. Plant products also play an important role in the healthcare systems of the remaining 20% of the population, mainly residents of developing countries. Medicinal plants are potential renewable natural resources. India is well known as an emporium of medicinal plants. Medicinal plants represent not only a valuable part of India's biodiversity but also a source of great knowledge. Geographically, India is located in the tropical zone. On account of its size; India is the home to a variety of environments from high snow capped mountain to tropical rain forest, hot to cold deserts, scrub lands to lush and fertile plains and valleys so also mangroves and seashores.

India has 16 agro climatic zones and medicinal plants are distributed across diverse habitats and landscapes. Nearly 70% of Indian medicinal plants are found in the tropical areas and other 30% of medicinal plants occur in temperate and Alpine, which include plants of highly medicinal values. Majority of medicinal plants are highly flowering plants. There are about 2200 genera which spreads over 386 families.

In India, Tamilnadu is under strategic geographical location and possesses an invaluable treasure of medicinal plants holding a major share in cultivation and export of medicinal plant species including *Sphaeranthes amaranthoides* (L.) Burm. Medicinal plants in Tamilnadu are cultivated in isolated patches each being grown in its favourable soil and agroclimatic conditions. Medicinal herbs are rich source of synthetic and herbal drugs. They contain a wide range of chemical compounds, commonly referred to as phytochemicals.

Modern agriculture emphasizes eco-friendly technologies such as organic farming and application of biofertilizers in crops. Current research in drug discovery from medicinal plants involves a multifaceted approach with the advent of innovative technologies and the importance being given to sustainable agriculture. Commercial cultivation of medicinal plants has started recently. In recent years due to over exploitation of natural resources, biofertilizers are emerged as an important component of integrated nutrient supply system (IPNSS) and hold a promise for reducing the production cost, improve the crop yields, quality nutrient supplies and sustaining the productivity over a longer period.

The rhizosphere is a habitat for a vast interactive community of rhizotrophic microorganisms whose activities largely determine the physico-chemical properties of the rhizosphere soil. A continuous interaction exists between the plant roots and the rhizotrophic microorganisms and within the different groups of these microorganisms which has an important influence on plant growth. In general, the population and microbial activity of rhizosphere is known to be several times higher than that of non-rhizosphere soil, because of higher nutrient availability in rhizosphere soil. The specific groups of microorganisms in rhizosphere such as nitrogen-Fixers, phosphorus solubilizers, Plant Growth Promoting Rhizomicroorganisms (PGPRs), Arbuscular Mycorrhizal Fungi (AMF) are considered beneficial, because their inoculation to soil is known to enhance nutrient content, biomass and yield of several plant species.

Of the various microorganisms colonizing the rhizosphere, mycorrhizae, the mutualistic symbionts, play an important role in mobilizing phosphorus from the

deeper layers of the soil and supplying it to the host plants. Among the mycorrhizae, Arbuscular Mycorrhizal Fungi (AMF) occupies a unique ecological position and is the most prevalent type. AMF represents one of nature's best gift to mankind in the conservation of arid soil to productive and fertile soil. AMF is a potent biofertilizer, nutrient remedifier, eco-friendly and used in agriculture, forestry and horticulture. AMF group has recently been erected to the status of a monophyletic phylum, the Glomeromycota. AMF is a unique symbiotic association between roots of higher plants and fungi. This association is of great economic significance in the growth of crop and medicinal plants.

AM fungi showed widespread occurrence in natural ecosystems. The increased interest in AM fungi in recent years has prompted numerous surveys, aimed at enumerating and assessing AM fungi in a particular habitat and plant species. Only scanty information is available on the correlation of edaphic characteristics to AM sporulation and colonization in the roots of medicinal plants under Indian conditions. Isolation and multiplication of native strains of AM fungi and other beneficial microbes are essential for mass production and tailoring of the roots of medicinal crops to make them suitable for growth in agricultural soils. The selection of an efficient strain of AM fungi is also an important step for the development of any successful mycorrhizal inoculation programme in medicinal crop plants. The significance of AM fungi in medicinal crop production is widely appreciated and therefore the necessity of using AM fungi and other efficient microbes as a bioinoculant for improving crop production is increasingly realized especially in transplantable medicinal crops.

The use of beneficial microbial inoculants including AM fungi and PGPRs play an important role in sustainable agriculture. But the research on AMF and PGPRs in medicinal plants is very limited. Some soil bacteria which have been named Mycorhriza Helper Bacterium (MHB), could enhance the development of the mycorrhizal symbiosis. It is well known that considerable number of bacterial species is also able to exert a beneficial effect of plant growth. Mostly they are associated with the plant rhizosphere; hence they are called "Rhizobacteria". This group of bacteria has been termed Plant Growth Promoting Rhizomicroorganisms (PGPRs), which were

discovered by Kloepper *et al.* (1991). PGPRs one of the great potential importance, their culture and mode of action are the subject of research. PGPRs are the synergistic effect with AMF in stimulating plant growth and also may stimulate root colonization by mycorrhizal fungi. Certain PGPRs have been reported to enhance activity of AM fungi and consequently plant growth. Therefore, microbial inoculants can help maintain good soil health and fertility that contribute to a greater extent to a sustainable yield and quality of products.

**(C) Definition of the Problem:**

Presently, considerable importance is being given to AM fungi and PGPRs, because of awareness of environmental pollution and health hazards by the use of chemicals. However, the information available on the use of AM fungi and PGPRs in medicinal plants is meager. Surprisingly, very little information on AM association with *Spharanthes amaranthoides* in tropical soils are available. However, for evoking better plant growth response and for enhancement of nutritional and phytochemical constituents of the inoculation of native AM fungi and PGPRs on *Spharanthes amaranthoides* is untapped resource and therefore, they merit study.

**(D) Scope of the research work:**

Hence, the present study was undertaken with the following objectives:

- to study the variability in infection rates, spore density and species richness of AM fungi associated with ***Sphaeranthes amaranthoides*** (L.) Burm. which was collected from two different localities of Thanjavur district, Tamil Nadu, India.
- to identify the efficient indigenous AM fungi and their mass multiplication in the roots of onion by pot culture for further use of the inoculation experiments.
- to study the effect of seven dominant native strains of AM fungi viz., *Archaeospora trappei, Acaulospora marrowae, Glomus pakistanika, Glomus walkeri, Gigaspora margarita* and *Scutellospora heterogama* on growth and nutrition of ***S. amaranthoides*** was determined by pot culture.
- to determine the morphometric parameters such as plant height, root and shoot biomass were recorded.

- To study the effect of *Glomus walkeri* (Wagaiyur isolate) and plant growth promoting rhizomicrobes *Bacillus subtilis* and *Trichoderma viride* on growth, nutrition and the content of secondary metabolites of *S. amaranthoides*.

**(E) Methodology**

- Soil samples were collected from the study sites and analysed for physico-chemical factors such as pH, organic carbon, EC, N, P, K, Zn, Mn, Cu, S and Fe (Jackson, 1973).
- Isolation of AM fungi from rhizosphere soils were assessed by wet-sieving and decanting technique (Gerdemann and Nicolson, 1963).
- Percent root colonization of AM fungi was determined by trypan blue lactophenol method (Phillips and Hayman, 1970).
- AM fungal species were identified by using manual for identification of AM fungi (Schenck and Perez, 1990).
- The bacterial cultures were obtained from Microbial Type Culture Collection (MTCC), Chandigarh, India.
- Disc diffusion method was employed for the study of antibacterial activity of the various solvent extracts (Onkarappa *et al.*, 2003).
- Sterile Muller Hinton Agar (MHA) was used.
- Control plates were prepared by adding dimethyl sulfoxide (DMSO) to separate discs.
- Statistical analysis was done by applying student's 't' test to find out the level of significance and tested for critical difference at 5% level of significance by DMRT.

**(F) Original contribution**

- Seven dominant indigenous AM fungi viz.,

***Archaeospora trappei,***

***Acaulospora marrowae,***

***Glomus aggregatum,***

***Glomus pakistanika,***

***Glomus walkeri,***

***Gigaspora margarita*** and

***Scutellospora heterogama***

were used for inoculum production.

- Isolation and mass multiplication of *Trichoderma* species and *Bacillus* species from rhizosphere soils of *S. amaranthoides*.
- to study the effect of seven dominant native strains of AM fungi viz., *Archaeospora trappei, Acaulospora marrowae, Glomus pakistanika, Glomus walkeri, Gigaspora margarita* and *Scutellospora heterogama* on growth and nutrition of ***S. amaranthoides*** was determined by pot culture.
- Among the seven indigenous AM fungi, ***Glomus walkeri*** was selected and performed best in improving plant growth and nutrition of ***S. amaranthoides.***
- to determine the morphometric parameters such as plant height, root and shoot biomass were recorded.
- The nutritional parameters such as N, P, K, Zn, Mn, Cu, S and Fe were analysed in the roots and leaves of ***S. amaranthoides***.
- To study the effect of *Glomus walkeri* (Wagaiyur isolate) and plant growth promoting rhizomicrobes *Bacillus subtilis* and *Trichoderma viride* on growth, nutrition and the content of secondary metabolites of *S. amaranthoides*.
- The following treatments were as follows:
    - Control
    - *Glomus walkeri* (Gw) alone
    - *Trichoderma viride* (Tv) alone
    - *Bacillus subtilis* (Bs) alone
    - Gw + Tv
    - Tw + Bs
    - Gw + Tv + Bc
- Qualitative analysis of phytochemical constituents such as, flavonoids, alkaloids, phytosterols, tannins, saponins, phenols and carbohydrate contents were determined in the mycorrhizal PGPRs and non-mycorrhizal treated leaves and roots of ***S. amaranthoides***.

- Quantitative analysis of enzymic and non-enzymic antioxidants such as acid and alkaline phosphatases, polyphenol oxidase and peroxidase were assayed in the mycorrhizal, PGPRs and non-mycorrhizal treated leaves and roots of ***S. amaranthoides***.
- To evaluate the presence of total flavonoids, alkaloids, tannins and phytosterols in the leaves and roots of the test plant.

**(G) Papers published : NA**

**(H) Conclusion:**

- The anti-bacterial activities of this plant for the treatments of diseases were also investigated in the present study.
- to determine the anti-bacterial activity of various solvent extracts of mycorrhizal, PGPRs and non-mycorrhizal leaves of ***S. amaranthoides*** against some common pathogenic bacterial organisms such as:

  Three Gram negative bacteria viz.,

  ***Escherichia coli,***

  ***Proteus vulgaris***

  ***Pseudomonas aeruginosa*** and

  two gram positive bacteria

  ***Staphylococcus aureus***

  ***Bacillus subtilis***

  were used as test organisms.
- The plates were incubated at 37°C for 24 h and the growth of inhibition zones were noted.
- The minimum inhibitory concentration of ***S. amaranthoides*** by using different bacterial cultures were also determined.
- ***S. amaranthoides*** mycorrhizal and PGPRs treated leaf extracts showed excellent inhibition zones against the specific tested pathogens namely ***S. aureus*** (16 mm), ***P. vulgaris*** (12 mm) and ***E. coli*** (10 mm).